

Title: Testing the uniqueness of *Z. h. intermedius* relative to *Z. h. campestris*

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Summary: The authors have submitted a comprehensive evaluation of the taxonomic status of Preble's Meadow Jumping Mouse. They have used an extensive array of data and associated analyses, including morphometric data and mtDNA data. They analyze these data in the context of a rigorous evaluation of both species status and ESU definitions. The authors conclude that there is no supporting evidence for the nominal subspecies. This conclusion is robust and strongly supported by all forms of data examined. It is not likely to be overruled by subsequent data collection. If populations were subsequently found to be distinct (genetically through microsatellite work or otherwise) it would clearly be the result of human-induced habitat fragmentation. The genetic data and morphometric data clearly support the idea that these populations were naturally and historically a single population with gene flow occurring on a regular basis between the subspecies that have been differentiated. The authors go on to evaluate that original subspecific definition in relation to these data. They clearly show that the subspecies in question was described **from three individuals** and the designation does not hold up to additional data. They justifiably synonymize *Z. h. preblei* and *Z. h. intermedius* with *Z. h. campestris*. Indeed, it is clear that the existing taxonomy for this whole genus is questionable and has been severely oversplit and that a comprehensive systematic study of the entire genus is in need to avoid such problems with taxonomy in the future.

Below I address the specific questions as requested:

1. The morphometric and phylogenetic methods used were appropriate. While there is some limitation in using just a single marker (D-loop mtDNA), the fact that there is very little variation here and that that fact is corroborated by a lack of morphological distinction is a powerful approach. The analytical methods employed could certainly have been improved in terms of more sophistication, but the results are robust to alternative methods. For example, the HKY model of evolution used is not justified at all. The Tajima test is a particularly weak test for deviations from neutrality. However, the MDIV analysis is quite well-done with a robust number of chains (5,000,000). The authors never actually perform an explicit test of their null hypothesis of the different subspecies forming monophyletic groups. That would be an improvement to the report. Such a test is easily done in PAUP* and would no doubt result in a significant rejection of the null hypothesis of subspecies monophyly.
2. The authors conclusions to synonymize the three subspecies is highly justified on both genetic and morphometric grounds. The data are clear and the original description of the subspecies was very weak.

3. Based on the MDIV analysis alone, it is clear that these two subspecies are not distinct. Including the morphometric and mtDNA phylogenetic data substantiates that conclusion even further.
4. The only alternative interpretation of the data is that there is ongoing gene flow due to human mediated activities or human induced corridors for gene flow. This alternative seems highly unlikely given the mtDNA phylogeny and the morphometric homogeneity.
5. I would recommend that the authors perform a nested clade analysis on the data to maximize the interpretive potential of the data. I would also suggest that, if the authors are willing and the agency interested in funding, the entire genus be examined using COI and cytb data. The genus is clearly a mess and the entire taxonomy can use an overhaul. Instead of D-loop, the authors should use the more conventional COI and cytb regions for easy comparisons with other studies.
6. No. The data were clear then and they are clear now. It is amazing that you are still spending time and resources on this (including my time and resources!).